



Title	Vertical position, specific gravity and swimming ability of Pacific cod <i>Gadus macrocephalus</i> yolk-sac larvae reared at four temperatures
Author(s)	Li, Zhe; Yamamoto, Jun; Sakurai, Yasunori
Citation	Fisheries science, 81(5), 883-889 https://doi.org/10.1007/s12562-015-0911-6
Issue Date	2015-09
Doc URL	https://hdl.handle.net/2115/62748
Rights	The final publication is available at Springer via http://dx.doi.org/10.1007/s12562-015-0911-6
Type	journal article
File Information	FS-Li Zhe.pdf



Title:

Vertical position, specific gravity and swimming ability of Pacific cod *Gadus macrocephalus* yolk-sac larvae reared at four temperatures

Authors:

Zhe Li¹, Jun Yamamoto², Yasunori Sakurai^{1*}

Author Affiliations :

¹ Graduate School of Fisheries Sciences, Hokkaido University, 3-1-1 Minato, Hakodate, Hokkaido 041-8611, Japan

² Field Science Center for Northern Biosphere, Hokkaido University, 3-1-1 Minato, Hakodate, Hokkaido 041-8611, Japan

***Corresponding author:** Yasunori Sakurai (sakurai@fish.hokudai.ac.jp)

Tel.: +81 0138 40 8861 / Fax: +81 138 8863

Authors E-mail: Zhe Li ; lz880526@live.cn

Jun Yamamoto ; yamaj@fish.hokudai.ac.jp

Abstract

Vertical position and movement rate were measured in the yolk-sac larvae of the Pacific cod *Gadus macrocephalus* at different rearing temperatures (3, 5, 7 and 9 °C) and an identical temperature of 5 °C in experimental columns during the initial 3 days post hatch (dph). Larval specific gravity was also measured through sinking velocities. Yolk-sac larvae swam upward and initially (0 dph) remained in the upper section of the experimental column through intermittent upward swimming and gradually moved to near-neutral buoyancy at 3 dph at corresponding rearing temperatures. Rearing temperature significantly affected the swimming ability: at 0 dph, vertical movement rates measured at 5 °C were low in the yolk-sac larvae reared at warm temperatures. However, no such difference was observed at 3 dph. The difference in vertical movement rate may have resulted from the temperature-mediated body sizes (standard length, SL) and yolk-sac sizes (yolk-sac volume, YSV) at hatching. Specific gravity did not change for the varying temperatures during the initial 3 days, but decreased as ontogeny progressed. Therefore, we suggest that Pacific cod yolk-sac larvae achieve a demersal-pelagic habitat transition through upward swimming and increasing buoyancy.

Key words: Pacific cod, yolk-sac larvae, various temperatures, vertical position, specific gravity, vertical movement rate

Introduction

Pacific cod *Gadus macrocephalus* is a commercially and ecologically important marine fish species that is widely distributed around the rim of the North Pacific Ocean [1, 2]. The females spawn demersal eggs from December to February [3, 4]. Pacific cod eggs remain strictly demersal during hatching [5]. After hatching, yolk-sac larvae actively swim upward as early as 1 day post hatch (dph) and tend to be near-neutrally buoyant until 3 dph, at which they appear to be negatively buoyant [6, 7]. In the environment of Mutsu Bay in Japan, yolk-sac larvae develop into pelagic larvae alongside a demersal-pelagic habitat transition [4, 8], which appears to be relatively rapid and associated with lipid/fatty acid synthesis [9]. Pacific cod larvae are thereby assumed to achieve vertical migration as a result of increased buoyancy just prior to hatching, rather than swimming [9]. However, the assumption is uncertain because the swimming ability of yolk-sac larvae may overcome negative buoyancy and migrate to surface layers [10]. On the other hand, yolk-sac larvae cannot swim continuously [11]; therefore, negatively buoyant larvae require a high swimming frequency to achieve the habitat transition and maintain their vertical position in surface layers [12]. In other words, both buoyancy and swimming behavior appear to play important roles in the habitat transition process.

Temperature potentially impacts larval size and endogenous reserves at hatching: warmer temperatures result in smaller body size but larger yolk-sac size [13, 14]. In addition, temperature affects physics of swimming conditions (e.g., dynamic viscosity) [15] and metabolic rates of fish larvae [16], both of which affect larval

67 swimming speed. Eventually, temperature has direct implications for larval survival
68 by regulating growth rate which is also related to body size [17-19], as well as
69 mortality through swimming performance that develops with increasing body size
70 [20-22].

71 The buoyancy of yolk-sac larvae is suggested to be highly related to egg density
72 and influences their vertical distributions [23]. Variation in specific gravity of
73 yolk-sac larvae may be due to asynchronous spawning and thus hatchings. Generally,
74 specific gravity changes with ontogeny, involving yolk absorption [23]; however,
75 such change differs among species: it increases in herring yolk-sac larvae with yolk
76 absorption, but decreases in Atlantic cod yolk-sac larvae [24].

77 Previous studies have mostly focused on horizontal swimming performance of fish
78 larvae [22, 25-28], while limited studies have described the vertical swimming
79 performance of fish larvae. Moreover, little is known regarding the demersal-pelagic
80 habitat transition of Pacific cod yolk-sac larvae.

81 The objectives of this study were to determine: (1) the vertical position of Pacific
82 cod yolk-sac larvae in an experimental column; (2) the effect of rearing temperature
83 on vertical movement rate at an identical temperature; and (3) change in specific
84 gravity of the yolk-sac larvae reared at various temperatures with age. Specific gravity
85 of warmer-reared Pacific cod larvae was expected to be higher due to larger yolk-sac
86 size at hatching [14, 24]. The relevance of these results is discussed in an attempt to
87 describe the physical process of the vertical migration of Pacific cod yolk-sac larvae.

88 **Materials and methods**

Egg collection and rearing

Adult Pacific cod were collected with bottom-set nets in Mutsu Bay, Aomori, Japan during the spawning season in February 2014. Gametes from one female and three males were mixed and rinsed at 4 °C. The fertilized eggs were shipped in an insulated container to the laboratory at the Faculty of Fisheries, Hokkaido University in Hakodate, Hokkaido, Japan. At about 5 h post-fertilization, the demersal eggs were scattered in a thin layer at the bottom of plastic containers (i.e., approximately 3,600 eggs/container, $N = 8$) within filtered seawater (4 °C, 34 in salinity) and two replicate containers were used for each of four rearing temperatures: 3, 5, 7 and 9 °C. Seawaters were aerated with air-stones to increase oxygen concentration and circulation to the demersal eggs in stagnant water. Time to hatch was defined as the day on which 50 % of the eggs had hatched at each temperature. At 0 dph of each rearing temperature, the yolk-sac larvae were transferred to polyethylene containers (400 ml) with air-bleed holes on the lids and reared at their incubation temperature for the initial 3 days, unfed but dependent on the yolk reserves. The rearing temperatures were 3.2 ± 0.4 , 5.2 ± 0.3 , 7.1 ± 0.2 and 8.9 ± 0.2 °C (mean $\pm$ SD) during the embryonic and larval period. Light regime was maintained at 12L:12D. Half of the seawater was replaced with fresh seawater at the corresponding temperature daily and every other day during the embryonic and larval period, respectively. Dead eggs or larvae were removed with a pipette.

Experimental design

Vertical position

Vertical positions of yolk-sac larvae were measured at their rearing temperatures at 0–3 dph (i.e., larvae reared at 3 °C were measured at 3 °C). An acrylic column (400 mm height, 60 mm diameter) was marked with 4-mm intervals on the outside and filled with chilled seawater at a specific temperature (i.e., 3, 5, 7 or 9 °C) to a depth of 320 mm. The trials were conducted in a temperature-controlled incubator with a transparent viewing window under a light intensity of about $6.8 \mu\text{E m}^{-2} \text{s}^{-1}$ at the water surface. Ten larvae at each temperature were gently and slowly introduced into the bottom of the acrylic column using a small column (40 mm height, 32 mm diameter) which was attached to a net lid and a slender tube. The net lid was gently opened by drawing a fastening string. After being allowed to freely swim for 10 min, vertical positions of the larvae were recorded through the transparent viewing window. Movement direction and angle of body orientation were additionally observed and estimated during swimming and resting bouts to determine whether the vertical position was maintained through buoyancy or swimming behavior [7, 10]. The seawater temperatures were measured before and after the observations with a thermometer (± 0.1 °C). Eight replicates were conducted in each rearing temperature/age combination.

Vertical movement rate

Vertical movement rates of yolk-sac larvae reared at the four temperatures were measured at an identical temperature of 5 °C at 0–3 dph. The identical temperature was used to eliminate the influence of ambient temperature on vertical movement rate, such as viscosity [15]. Prior to each replicate, twenty larvae were acclimated at 5 °C

for 15 min except those reared at 5 °C. The larvae were then transferred into the bottom of the acrylic column in the same way as in the vertical position trials. To minimize repeated recordings as far as possible, only ten of the twenty larvae were randomly selected and recorded using a video camera (Sony HDR-CX370) through the transparent viewing window. In addition, movement distance less than 4 mm was not considered since the behavior may be due to twisting, rather than swimming [29]. The seawater temperatures were also measured before and after the recordings with the same thermometer. A total of 13–36 larvae from eight replicates were subsampled and photographed for measuring the standard length (SL) and yolk-sac volume (YSV) in each rearing temperature/age combination.

Specific gravity

The experimental method generally followed that of Neilson et al. [30] on larval Atlantic cod *Gadus morhua*. Specific gravities of Pacific cod yolk-sac larvae were determined by measuring sinking velocities in seawaters of four density types. Four acrylic columns (400 mm height, 65 mm diameter) were marked at 5-cm intervals and filled with filtered seawater of known salinities to a depth of 350 mm. The seawater salinities were measured with a YSI EC300 (with a resolution of 0.002) and adjusted to 10, 15, 20 and 25 with artificial sea salt (Rohto marine, Rei-Sea, Tokyo, Japan). An Onset HOBO® Pendant Temperature logger UA-001-08 (Onset Computer, Bourne, HOBO, Massachusetts, USA) was used to note the temperatures during the measurement period. Seawater densities were calculated with the known salinities and noted temperatures [31].

At a fixed time, twenty larvae in each rearing temperature/age combination were individually anesthetized in FA100 (Dainippon Sumitomo Pharma, Japan) for approximately 1 min and after rinsed in filtered seawater with a certain test salinity ($N = 4$), they were gently released at the surface of the seawater with the corresponding salinity. The time to sink 10 cm was measured with a resolution of 0.01 s after sinking 5 cm to obtain a stable speed. The anesthetic time was short and the larvae were seemingly not influenced by the anesthetic, since all of them revived after the trials.

Data analysis

In each rearing temperature/age combination, the median of vertical positions was calculated in eight replicates and regarded as the level of vertical position. The mean 25th and 75th percentiles of vertical positions were also calculated to describe the vertical position range of the yolk-sac larvae in the experimental column. Two-way ANOVA was employed to examine the difference in vertical positions in the experimental column with rearing temperature ($N = 4$) and age ($N = 4$) as the main factors.

The vertical movement rate was defined as vertical movement distance divided by time (mm/s). Video analysis software (MEPG video Wizard DVD) was used to calculate vertical movement rates at 1/30 s accuracy. At each age, the Kruskal-Wallis rank-sum test was used to determine the difference in vertical movement rate across the various rearing temperatures. Pairwise multiple comparisons were conducted using the Kruskal-Wallis test (function 'Kruskalmc' in the R package 'pgirmess') within R version 3.1.2. A linear regression model ($y = ax + b$) was used to clarify the

relationships between vertical movement rate and temperature-specific morphometric characteristics: body size (SL) and yolk-sac size (YSV) measured from all the sampled larvae [32].

At each salinity used for testing, 20 sinking velocity measurements were grouped into four sections (i.e., 5 measurements/section). The mean sinking velocity in each section was treated as one replicate to a particular salinity (i.e., four replicates/salinity). A linear regression ($\rho = a + b\omega$) or an exponential regression model ($\rho = a\omega^b$) was used to describe the relationship between sinking velocities (ω) and seawater densities (ρ) (i.e., one replicate sinking velocity/salinity). The chosen regression model was based on the higher regression coefficient. The point at which $\omega = 0$, which was referred as neutral buoyancy, was assumed to represent larval specific gravity to this section and back-calculated from the determined regression model. The mean specific gravity from four sections was regarded as the specific gravity in a rearing temperature/age combination. The change in the larval specific gravity was analyzed with the linear regression model ($y = ax + b$). The differences in specific gravity for various temperatures were examined using the Kruskal-Wallis rank-sum test across the ages. Statistical significance was set at $p < 0.05$.

Results

The yolk-sac larvae swam upward and maintained position at the upper half of the experimental column as early as 0 dph at all the rearing temperatures (Table 1; Fig. 1). Neither rearing temperature nor age significantly affected vertical position during the initial 3 days (Fig. 1; $p > 0.05$).

Swimming behavior markedly changed during the initial 3 days. At 0 dph, yolk-sac larvae swam upward in nearly straight routes and sank quickly at an angle of almost 90° (degree from horizontal) during swimming and resting bouts, respectively. The swimming behavior utilized can be described as burst swimming. At 3 dph, yolk-sac larvae swam upward in sinuous routes and sank relatively slowly at an angle of 30° – 45° during swimming and resting bouts, respectively. The swimming behavior utilized was similar to the routine swimming. In general, yolk-sac larvae tended to be negatively buoyant and maintained vertical positions by intermittent swimming at 0 dph. As ontogeny progressed, yolk-sac larvae maintained vertical positions by their near-neutral buoyancy instead of swimming at 3 dph. Some larvae stayed at the bottom during the observation period (Supplementary video). All the mentioned swimming behaviors were consistently observed at all the rearing temperatures.

At an identical temperature of 5 °C (Table 1), vertical movement rate remained lowest in the larvae reared at 9 °C at 0–2 dph, and was lower in the larvae reared at 7 °C than that at 3 °C only at 0 dph (Fig. 2; $p < 0.05$). At 3 dph, there was no significant difference in vertical movement rates of larvae reared at the various temperatures (Fig. 2; $p > 0.05$). In the current study, vertical movement rate was positively correlated with body size (Fig. 3; $p < 0.001$) and negatively correlated with yolk-sac size (Fig. 4; $p < 0.01$) across the rearing temperatures during the initial 3 days. Subsampled 21–79 individuals were taken into account in each rearing temperature/age combination (Table 2).

In the current study, Pacific cod yolk-sac larvae were negatively buoyant relative to

the densities of the seawaters (1.0258 (B)–1.0265 (A) g/cm³ at 3–9 °C, Fig. 5) at 0 dph. The specific gravity of yolk-sac larvae decreased with age at all the rearing temperatures (Fig. 5; all $p < 0.05$). No consistent differences in specific gravities were observed for the temperatures across the initial 3 days. At 3 dph, however, all specific gravities decreased to the level close to the seawater densities (Fig. 5), indicating that the yolk-sac larvae were near-neutrally buoyant at this stage.

Discussion

Three laboratory experiments were conducted to determine the method that Pacific cod yolk-sac larvae utilize to migrate upward and whether rearing temperature affects the vertical movement ability of yolk-sac larvae. In the current study, the yolk-sac larvae swam upward as early as 0 dph and the vertical position did not change across the rearing temperatures or ages. These results are similar to the previous study by Hurst et al. [7] reporting that yolk-sac larvae swim upward and remain at an upper position as early as 1 dph at 4 °C. Since first feeding occurs at depletion of approximately 50 % yolk reserves [33], vertical migration of pre-feeding larvae appears to coincide with first feeding. In the Baltic Sea, *Gadus morhua* yolk-sac larvae have been thought to reach the upper water layers where prey availability is high as soon as possible, at least before the yolk reserves are completely depleted [12, 33]. In the environment of Mutsu Bay, Pacific cod yolk-sac and yolk-absorbed larvae mainly inhabit depths at 8–45 m where their favorable prey, copepod nauplii, is abundant [4].

243 Compared with the field depth, the experimental column depth in the current study
244 was extremely small (i.e., 32 cm), possibly allowing the yolk-sac larvae to reach the
245 upper half of the experiment column in 10 min. Therefore, there was no difference in
246 the vertical positions within the temperatures or ages in the current study. However,
247 the different strategies in which larvae maintain vertical position suggest that larval
248 specific gravity decreases with age. As a result, larvae do not require high swimming
249 activity to maintain vertical position, but depend rather on neutral buoyancy. The
250 observation of some individuals remaining at the bottom of the acrylic column may be
251 due to their periodically resting there [7] or swimming upward after the observations.

252 Vertical movement rate of larvae reared under warmer conditions was initially
253 low at an identical temperature of 5 °C. This might have resulted from the
254 temperature-mediated body sizes and yolk-sac sizes at hatching [14, 32]. At 3 dph, the
255 convergence in vertical movement rate may be due to the same ambient temperature
256 (5 °C) rather than the morphometric differences.

257 The specific gravity of the yolk-sac larvae generally decreased with age during the
258 initial 3 days (Fig. 5). The result was affirmed by the observations that the larvae sank
259 quickly during resting bouts at 0 dph, but slowly at 3 dph. Moreover, Laurel et al. [9]
260 proposed that Pacific cod larvae achieve vertical migration through a rapid decrease in
261 specific gravity. Similarly, North Sea herring larvae are also negatively buoyant at
262 hatching and migrate upward, involving yolk absorption and decreasing specific
263 gravity. Decreased specific gravity appears to be a physiological adjustment to
264 facilitate the attainment of surface layers for first feeding and energetic conservation

265 because of low first feeding success [24]. Although specific gravity was expected to
266 be positively related with temperature at hatching, the homogenous specific gravities
267 might have resulted from temperature-specific development rates during time delays.
268 Specific gravities were not measured until the point of 50 % hatching, during which
269 the yolk-sac larvae underwent temperature-specific decreases in specific gravity. After
270 measurements, all the larvae possibly achieved a similar specific gravity.

271 From the above considerations, vertical migration of Pacific cod yolk-sac larvae
272 appears to be associated with upward swimming and increasing buoyancy. Upward
273 swimming ensures that the yolk-sac larvae depart from the bottom water layers. They
274 exhibited active and intermittent swimming behavior which initially offsets the
275 negative buoyancy. However, yolk-sac larvae may immediately descend during
276 resting bouts due to negative buoyancy and possibly hover within a specific depth in
277 the field. Thus initial net upward movement was hardly or limitedly achieved. In this
278 case, since both swimming ability and buoyancy increase as ontogeny progresses,
279 larvae swim upward rather easier or even float without swimming [9]. Increased
280 buoyancy may gradually replace swimming behavior as the dominant factor to
281 maintain vertical position in this species. Thereafter, larvae might be capable of
282 selecting vertical positions to some extent by swimming behavior and maintain their
283 positions by neutral buoyancy.

284 Interestingly, the result of change in buoyancy contrasts with the previous study of
285 Hurst et al. [7] demonstrating that Pacific cod larvae are neutrally to negatively
286 buoyant over the period from hatching to 3 dph. The divergence between the results

may be due to population-specific physiological adaptations to different habitats. The different depths of spawning locations, for example, may lead to different strategies of vertical migration to surface layers. The spawning location is deeper in the Gulf of Alaska (> 100 m) than that in Mutsu Bay (about 60 m) [4, 9]. Yolk-sac larvae thus have to migrate upward further in the Gulf of Alaska [4, 34]. Consequently, the larvae may complete vertical migration sooner by neutral buoyancy at hatching in the Gulf of Alaska, and maintain vertical position by intermittent swimming as a decrease in buoyancy occurs. On the other hand, larvae in Mutsu Bay may not need to migrate upward so early as those in the Gulf of Alaska. The larvae may hover for a period due to weak swimming ability and high specific gravity, and become capable of ascending to surface layers in a short time as an increase in buoyancy occurs [9].

Ecological implications

The demersal-pelagic habitat transition is an essential physiological process for Pacific cod yolk-sac larvae after hatching, inducing yolk-sac larvae to depart from the low oxygen zone at hatching depths and initiate first feeding at depths where favorable prey availability is expected [12] in the method involving upward swimming and increasing buoyancy. If some larvae fail to achieve the habitat transition before first feeding or yolk depletion, they are likely to suffer high mortality due to low oxygen concentration and prey availability [12]. Thus, this process appears to play an important role in larval survival and even recruitment during their early life stages [35].

The temperature-mediated swimming ability is likely to be an adaptive strategy to

various spawning environments. Larvae hatching in a low temperature obtain high swimming ability that might offset the effects of low metabolic rate and high viscosity in the low temperature [15]. However, it is unclear that how far larval swimming ability offsets the impacts of a low water temperature. Furthermore, whether (and how) water temperature impacts the habitat transition speed of this species is still unknown. If water temperature impacts more than larval swimming ability, larvae hatching in a higher temperature are likely to complete the habitat transition and start first feeding earlier than those hatching in a lower temperature. Meanwhile, an earlier habitat transition might be harmful to yolk-sac larvae because they may be exposed to predators earlier and high mortality is consequently expected. In this case, water temperature, influencing the habitat transition speed, appears to become an indicator of survival of Pacific cod yolk-sac larvae. Therefore, further studies are required to clarify the role of water temperature during the demersal-pelagic habitat transition of Pacific cod yolk-sac larvae.

Acknowledgments

We thank to the Wakinosawa Fishery Association for offering great quality Pacific cod eggs. Thanks are also extended to two anonymous reviewers for detailed revision and helpful comments on this manuscript.

References

1. Alderdice D, Forrester C (1971) Effects of salinity, temperature, and dissolved oxygen on early development of the Pacific cod (*Gadus macrocephalus*). J Fish Res Board Can 28: 883-902
2. Laurel B, Stoner A, Ryer C, Hurst T, Abookire A (2007) Comparative habitat associations in juvenile Pacific cod and other gadids using seines, baited cameras and laboratory techniques. J Exp Mar Biol Ecol 351: 42-55
3. Sakurai Y, Hattori T (1996) Reproductive behavior of Pacific cod in captivity. Fish Sci 62: 222-228
4. Takatsu T, Nakatani T, Miyamoto T, Kooka K, Takahashi T (2002) Spatial distribution and feeding habits of Pacific cod (*Gadus macrocephalus*) larvae in Mutsu Bay, Japan. Fish Oceanogr 11: 90-101
5. Bian X, Zhang X, Sakurai Y, Jin X, Gao T, Wan R, Yamamoto J (2014) Envelope surface ultrastructure and specific gravity of artificially fertilized Pacific cod *Gadus macrocephalus* eggs. J Fish Biol 84: 403-421
6. Forrester C (1964) Laboratory observations on embryonic development and larvae of the Pacific cod (*Gadus macrocephalus* Tilesius). J Fish Res Board Can 21: 9-16
7. Hurst TP, Cooper DW, Scheingross JS, Seale EM, Laurel BJ, Spencer ML (2009) Effects of ontogeny, temperature, and light on vertical movements of larval Pacific cod (*Gadus macrocephalus*). Fish Oceanogr 18: 301-311
8. Sakurai Y (2007) An overview of the Oyashio ecosystem. Deep-Sea Res Pt II 54: 2526-2542
9. Laurel BJ, Copeman LA, Hurst TP, Parrish CC (2010) The ecological significance of lipid/fatty acid synthesis in developing eggs and newly hatched larvae of Pacific cod (*Gadus macrocephalus*). Mar Biol 157: 1713-1724
10. Davis MW, Olla BL (1994) Ontogenetic shift in geotaxis for walleye pollock, *Theragra chalcogramma* free embryos and larvae: potential role in controlling vertical distribution. Environ Biol Fishes 39: 313-318
11. Webb PW, Weihs D (1986) Functional locomotor morphology of early life-history stages of fishes. Trans Am Fish Soc 115: 115-127
12. Grønkjær P, Wieland K (1997) Ontogenetic and environmental effects on vertical distribution of cod larvae in the Bornholm Basin, Baltic Sea. Mar Ecol Prog Ser 154: 91-105
13. Methven DA, Brown JA (1991) Time of hatching affects development, size, yolk volume, and mortality of newly hatched *Macrozoarces americanus* (Pisces: Zoarcidae). Can J Zool 69: 2161-2167
14. Laurel BJ, Hurst TP, Copeman LA, Davis MW (2008) The role of temperature on the growth and survival of early and late hatching Pacific cod larvae (*Gadus macrocephalus*). J Plankton Res 30: 1051-1060
15. Hunt von Herbing I (2002) Effects of temperature on larval fish swimming performance: the importance of physics to physiology. J Fish Biol 61: 865-876
16. Beveridge OS, Petchey OL, Humphries S (2010) Mechanisms of temperature-dependent swimming: the importance of physics, physiology and

- body size in determining protist swimming speed. J Exp Biol 213: 4223-4231
17. Pepin P (1991) Effect of temperature and size on development, mortality, and survival rates of the pelagic early life history stages of marine fish. Can J Fish Aquat Sci 48: 503-518
 18. Otterlei E, Nyhammer G, Folkvord A, Stefansson SO (1999) Temperature- and size-dependent growth of larval and early juvenile Atlantic cod (*Gadus morhua*): a comparative study of Norwegian coastal cod and northeast Arctic cod. Can J Fish Aquat Sci 56: 2099-2111
 19. Folkvord A (2005) Comparison of size-at-age of larval Atlantic cod (*Gadus morhua*) from different populations based on size- and temperature-dependent growth models. Can J Fish Aquat Sci 62: 1037-1052
 20. Litvak MK, Leggett WC (1992) Age and size-selective predation on larval fishes - the bigger-is-better hypothesis revisited. Mar Ecol Prog Ser 81: 13-24
 21. Leggett W, Deblois E (1994) Recruitment in marine fishes: is it regulated by starvation and predation in the egg and larval stages? Neth J Sea Res 32: 119-134
 22. Shepherd T, Costain K, Litvak M (2000) Effect of development rate on the swimming, escape responses, and morphology of yolk-sac stage larval American plaice, *Hippoglossoides platessoides*. Mar Biol 137: 737-745
 23. Saborido-Rey F, Kjesbu OS, Thorsen A (2003) Buoyancy of Atlantic cod larvae in relation to developmental stage and maternal influences. J Plankton Res 25: 291-307
 24. Yin M, Blaxter J (1987) Temperature, salinity tolerance, and buoyancy during early development and starvation of Clyde and North Sea herring, cod, and flounder larvae. J Exp Mar Biol Ecol 107: 279-290
 25. Batty R, Blaxter J (1992) The effect of temperature on the burst swimming performance of fish larvae. J Exp Biol 170: 187-201
 26. Childs MR, Clarkson RW (1996) Temperature effects on swimming performance of larval and juvenile Colorado squawfish: implications for survival and species recovery. Trans Am Fish Soc 125: 940-947
 27. Green BS, Fisher R (2004) Temperature influences swimming speed, growth and larval duration in coral reef fish larvae. J Exp Mar Biol Ecol 299: 115-132
 28. Peck MA, Buckley LJ, Bengtson DA (2006) Effects of temperature and body size on the swimming speed of larval and juvenile Atlantic cod (*Gadus morhua*): implications for individual-based modelling. Environ Biol Fishes 75: 419-429
 29. Døving K, Mørstøl M, Andersen J, Knutsen J (1994) Experimental evidence of chemokinesis in newly hatched cod larvae (*Gadus morhua* L.). Mar Biol 120: 351-358
 30. Neilson JD, Perry RI, Valerio P, Waiwood KG (1986) Condition of Atlantic cod *Gadus morhua* larvae after the transition to exogenous feeding - morphometrics, buoyancy and predator avoidance. Mar Ecol Prog Ser 32: 229-235
 31. Millero FJ, Poisson A (1981) International one-atmosphere equation of state of

417 seawater. Deep-Sea Res 28: 625-629

418 32. Bian X, Zhang X, Sakrai Y, Jin X, Gao T, Wan R, Yamamoto J (2014)

419 Temperature - mediated survival, development and hatching variation of

420 Pacific cod *Gadus macrocephalus* eggs. J Fish Biol 84: 85-105

421 33. Hunt von Herbing I, Boutilier R, Miyake T, Hall B (1996) Effects of

422 temperature on morphological landmarks critical to growth and survival in

423 larval Atlantic cod (*Gadus morhua*). Mar Biol 124: 593-606

424 34. Boehlert GW, Gadomski DM, Mundy BC (1985) Vertical distribution of

425 ichthyoplankton off the Oregon coast in spring and summer months. Fish Bull

426 83: 611-621

427 35. Houde E, Hoyt R (1987) Fish early life dynamics and recruitment variability.

428 Am Fish Soc Symp 2: 17-29

429

430

431

Figure captions and Table titles

Fig. 1 Vertical position of Pacific cod larvae in the experimental column during the initial 3 days after hatching at 3 (**a**), 5 (**b**), 7 (**c**) and 9 °C (**d**). Data points represent the average of median position in eight replicates and error bars present average 25th and 75th percentiles of larval positions

Fig. 2 Mean vertical movement rate of Pacific cod larvae at 5 °C during the initial 3 days after hatching. The larvae were reared at 3, 5, 7 and 9 °C, respectively. Data points and error bars represent means $\pm$ SE calculated from 21–79 individuals in eight replicates

Fig. 3 Relationship between vertical movement rate (V) and body size (standard length, SL) of larvae reared at 3, 5, 7 and 9 °C during the initial 3 days after hatching

Fig. 4 Relationship between vertical movement rate (V) and yolk-sac size (volume, YSV) of the larvae reared at 3, 5, 7 and 9 °C during the initial 3 days after hatching. The data were multiplied by 1,000 at the x-axis

Fig. 5 Specific gravity of Pacific cod larvae reared 3, 5, 7 and 9 °C during the initial 3 days after hatching. Line A and B stand for seawater densities at 3 and 9 °C, respectively. Data points and error bars represent means $\pm$ SD

Table 1 Actual temperatures (mean $\pm$ SD, °C) in trials of vertical position (A) and vertical movement rate (B)

Table 2 Standard length (SL, mean $\pm$ SD) and yolk-sac volume (YSV, mean $\pm$ SD) of larvae reared at 3, 5, 7 and 9 °C during the initial 3 days after hatching, based on 13–36 subsample individuals at each temperature/age combination

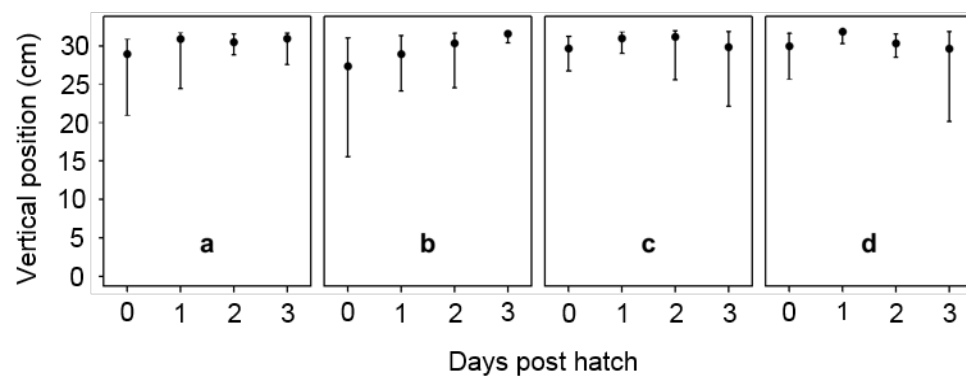


Fig. 1

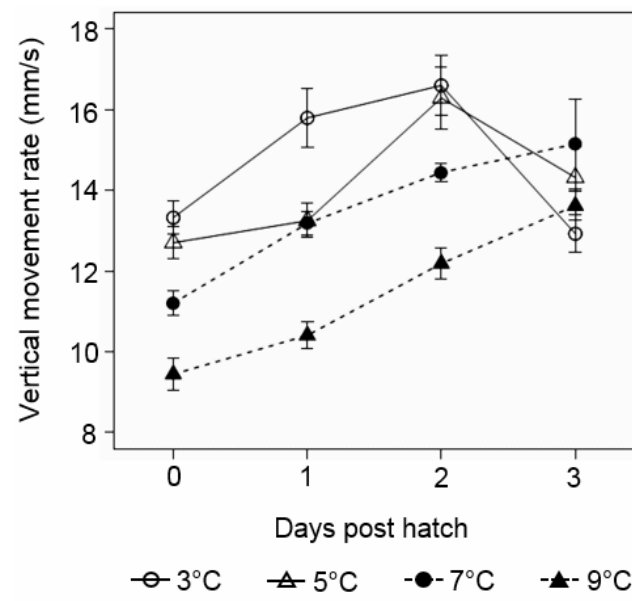


Fig. 2

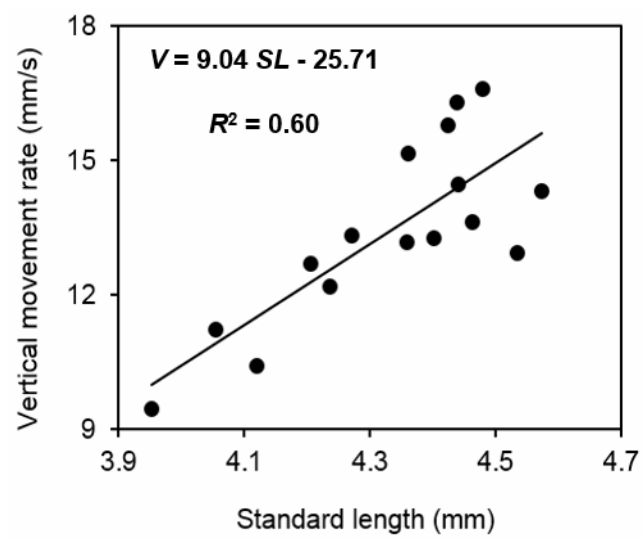


Fig. 3

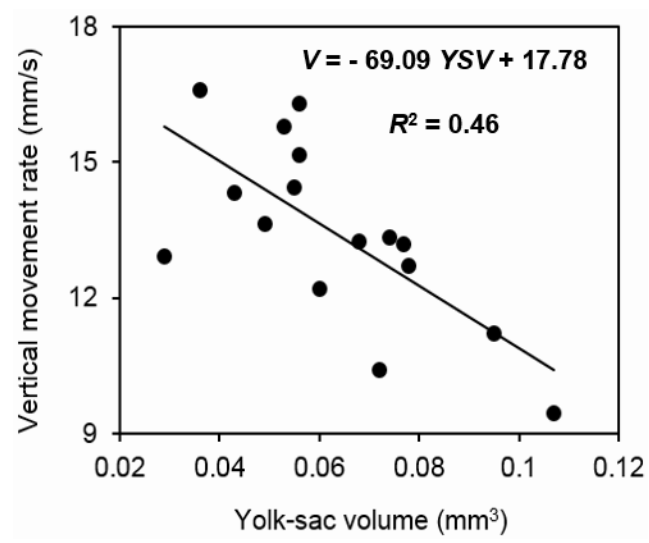


Fig. 4

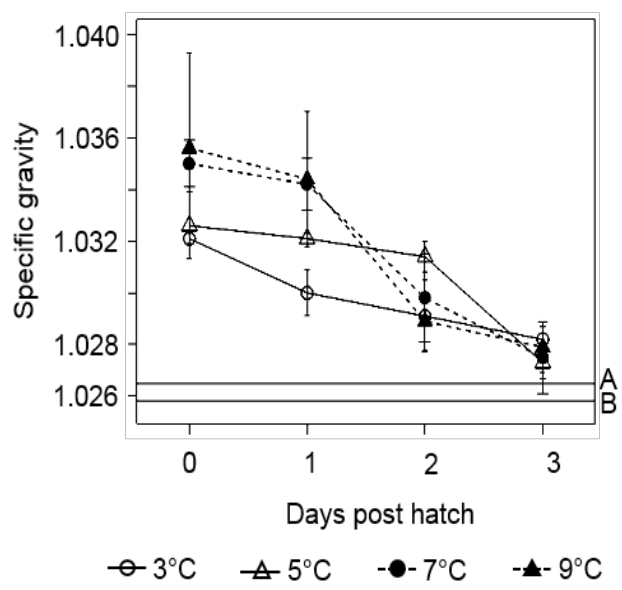


Fig. 5

Table 1

Temperature treatment (°C)	Actual temperature (mean $\pm$ SD, °C)	
	A	B
3	3.2 $\pm$ 0.2	5.1 $\pm$ 0.2
5	5.1 $\pm$ 0.1	5.1 $\pm$ 0.1
7	7.1 $\pm$ 0.2	5.2 $\pm$ 0.2
9	9.0 $\pm$ 0.3	5.3 $\pm$ 0.1

Table 2

Temperature (°C)	Days post hatch					
	0		1		2	
	SL (mm)	YSV (mm ³)	SL (mm)	YSV (mm ³)	SL (mm)	YSV (mm ³)
3	4.27±0.16	0.074±0.022	4.42±0.09	0.053±0.013	4.48±0.13	0.036±0.007
5	4.21±0.09	0.078±0.033	4.40±0.11	0.068±0.011	4.44±0.13	0.056±0.012
7	4.05±0.19	0.095±0.027	4.36±0.18	0.077±0.019	4.44±0.14	0.055±0.014
9	3.95±0.10	0.107±0.029	4.12±0.12	0.072±0.014	4.24±0.20	0.060±0.011
					4.46±0.13	0.049±0.009